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INVESTIGATIONS INTO THE MICROBIAL ECOLOGY AND LIMNOLOGY
OF HYRUM RESERVOIR, IN NORTHERN UTAH

by

Kenneth Maxwell Green

A thesis submitted in partial fulfillment
of the requirements for the degree

of

MASTER OF SCIENCE

in

Bacteriology and Public Health

Approved:

UTAH STATE UNIVERSITY
Logan, Utah

1971

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ABSTRACT

Investigations into the Microbial Ecology and Limnology
of Hyrum Reservoir, in Northern Utah

by

Kenneth Maxwell Green, Master of Science

Utah State University, 1971

Major Professor: Dr. Frederick J. Post
Department: Bacteriology and Public Health

A series of preliminary investigations was carried out to determine the factors promoting the dense, late summer waterblooms of Aphanizomenon flos-aquae in Hyrum Reservoir in northern Utah. Attempts were made to culture the Aphanizomenon in the ASM-8a medium of O'Flaherty and Phinney (J. Phycol. 6:95-97. 1970), but no growth was obtained and the algae soon lysed. Cultures were maintained, without growth, for more than four months in a lake water--lake sediment medium at 17 C under 1500 lux fluorescent light.

Dissolved oxygen measurements using an in situ probe revealed the development of a sharp oxycline during the summer months; on one day the dissolved oxygen concentration was observed to drop from 118% saturation (8.6 mg O₂/l) at 6.5 meters depth, to 33% saturation (2.4 mg O₂/l) at 7 meters, with a concurrent temperature decrease from 21.5 C to 21 C.

During the same period, pH was found to drop from 8.6 at the surface to 8.5 at 5 meters, 8.0 at 10 meters, and to 7.8 at the

bottom (16 meters). Secchi disc depth corresponded to 14% of the incident radiation, this depth varying from 1.3 to 1.8 meters during the algal bloom. The photic zone (1% of incident radiation) extended to 3.5 meters depth.

Water samples were collected from late April until early October, and these were analyzed for total organic carbon (TOC) using a Beckman model 915 total organic carbon analyzer. The organic carbon concentrations were found not to vary significantly with season or location. Many of the samples contained large numbers of Aphanizomenon but carbon analyses did not reflect this. It was concluded that the phytoplankton carbon in the reservoir was so much smaller than the carbon in the form of other organic materials, such as microseston, bacteria, detritus, colloids, and dissolved material, that fluctuations in algal carbon were therefore masked by the large amount of carbon continually present in these other forms. The mean organic carbon concentration for the lake was 4.6 mg/l (n=118, s=1.47) and the range was from 1.2 to 8.9 mg TOC/l.

The repeatability of measurements with the carbon analyzer is only within a range of 2 mg C/l, so the instrument is not sufficiently accurate for lake water analysis without the use of concentration techniques. Some trends were observed, but only at a low level of statistical significance: TOC concentration decreased slightly with depth, and TOC was correlated with stream discharge in the Little Bear River, which feeds the reservoir. An increase in TOC concentration in the reservoir during the algal bloom could not be verified statistically.

INTRODUCTION

Hyrum Reservoir, in Cache County, northern Utah, although originally constructed in 1935 for storage of irrigation water, has since assumed great importance as a recreation area. It has been incorporated into Hyrum Lake State Park and is used now for fishing, boating, water skiing, and swimming. Unfortunately, the fish population is affected by a copepod ectoparasite, Lernaea cyprinacea, commonly known as "anchor worm" (Rich, 1960). In August and September an intense bloom of Aphanizomenon flos-aquae, a filamentous blue-green alga, makes the water unattractive for swimming.

The current investigation is a preliminary part of a study to determine the causes of the algal bloom. The major algae in the lake have been identified and the Aphanizomenon cultured in the laboratory. Dissolved oxygen, temperature, light penetration, pH, and Secchi disc measurements were carried out on the site, and water samples were collected and analyzed for total organic carbon concentration. The data provided by these studies are being used to give direction to future investigations, leading ultimately to solution of the bloom problem.

Acknowledgment is made of the assistance provided by Dr. Raymond I. Lynn and Dr. Donald B. Porcella during these investigations. The project is being supported by the Office of Water Resources Research (CWRR-19), and the investigator has been supported throughout his Master's program by a Federal Water Quality Administration (now Environmental Protection Agency) traineeship.

HYRUM RESERVOIR

Hyrum Reservoir is located at latitude $41^{\circ}37'30''$, longitude $111^{\circ}52'30''$, in SE $\frac{1}{4}$ NE $\frac{1}{4}$ sec. 7, T.10 N., R.1 E., Cache County, Utah, 1 mile southwest of the town of Hyrum. It is fed by the Little Bear River, which, with its tributaries, represents a drainage area of 570 sq km (220 sq mi). The reservoir is formed by an earth-fill dam; storage began in 1935; its total capacity is 24.2 million cu m (19,600 acre-ft), of which the lower 4.2 million cu m (3,400 acre-ft) is below the sill of the outlet canal and is therefore considered dead storage. In 1970 the maximum content observed was 24.1 million cu m (19,500 acre-ft), on May 21, and the minimum occurred on September 5—14.3 million cu m (11,600 acre-ft) (USDI, 1971). This represents a drop in water level of about 5.5 meters (18 feet). The maximum and minimum depths observed at the deepest part of the reservoir during the present investigation were 21 meters (June 14) and 16 meters (August 28), respectively.

Using data provided by the U.S. Geological Survey (USDI, 1971) and the depths recorded on the various sampling days, a graph of water surface elevation and depth at station D, the deepest part of the reservoir, versus usable contents and total capacity has been drawn (Figure 1). By use of this graph it is possible to determine the volume of water in the reservoir by measuring the depth at station D. The graph has also been used to calculate that approximately 1.5 meters of sediment has accumulated at station D since the reservoir was built

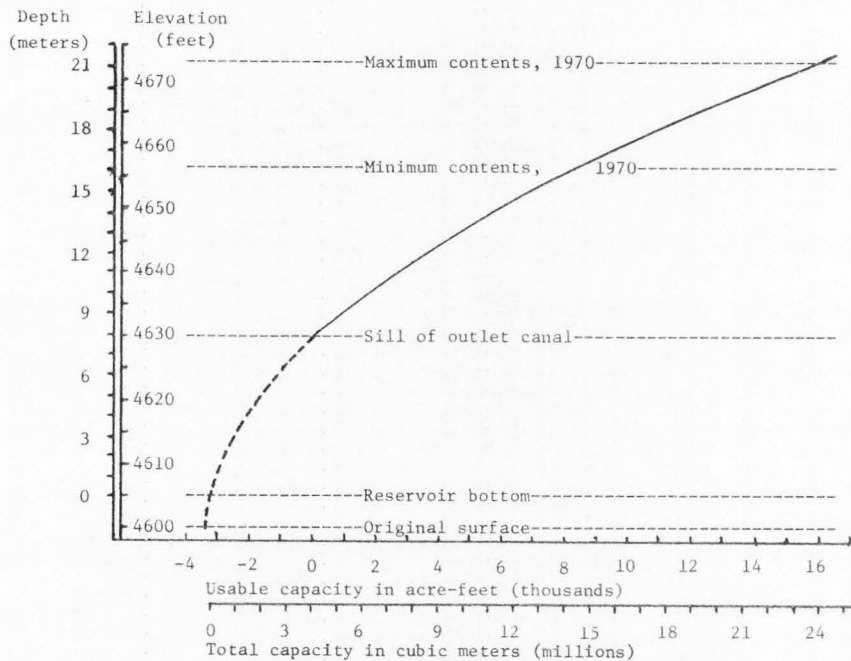


Figure 1. Water surface elevation and depth at station D versus usable contents and total capacity.

in 1935. This is between four and five centimeters of sediment accumulation per year. Also determined from the graph is that the sill of the outlet canal is 7.6 meters above the sediment surface. Because the dotted lines in the graph delineating the "dead storage" area and the sediment surface are extrapolations based on field measurements and old maps (USDI, 1926), the above conclusions must be regarded as tentative.

Sampling stations (Table 1 and Figure 2) were chosen with an eye to obtaining the most information from as small a number of stations as possible. Accessibility was also an important factor in station selection. Two stations, designated A and C, were located along the Little Bear River, which feeds Hyrum Reservoir. Each was accessible by automobile, and samples were collected by lowering a container from the bridge which crosses the river. Station A is upstream from White's Trout Farm, the effluent from which flows into Little Bear River, and station C is located just below the trout farm. Station B samples trout farm effluent directly.

Station D is located at the outlet to Hyrum Reservoir, and can be sampled directly from the bridge over the spillway or by boat. This is the deepest part of the reservoir, and the location was marked for boat sampling by a Park Service buoy anchored just in front of the spillway outlet. With care, it was possible to secure the boat to the buoy without falling into the lake. The position was then maintained while measurements or samples were taken.

Stations A, B, C, and D were considered the most important. Samples were also taken, but less frequently, from the area draining

Table 1. Sampling locations

Station designation	Site description
A	Little Bear River above White's Trout Farm.
B	Effluent from White's Trout Farm.
C	Little Bear River below White's Trout Farm.
D	Hyrum Reservoir outlet. Deepest part of reservoir.
E	Hyrum Reservoir at source of influent from Hyrum town dump.
F	Hyrum Reservoir geometric center.
G	Hyrum Reservoir swimming area.
H	Hyrum Reservoir near inflow of Little Bear River.
I	Little Bear River near entrance to Hyrum Reservoir.
P	Porcupine Reservoir; a new, unpolluted reservoir upstream from Hyrum Reservoir.
Bloom	Dense algal patch. Location not determined.

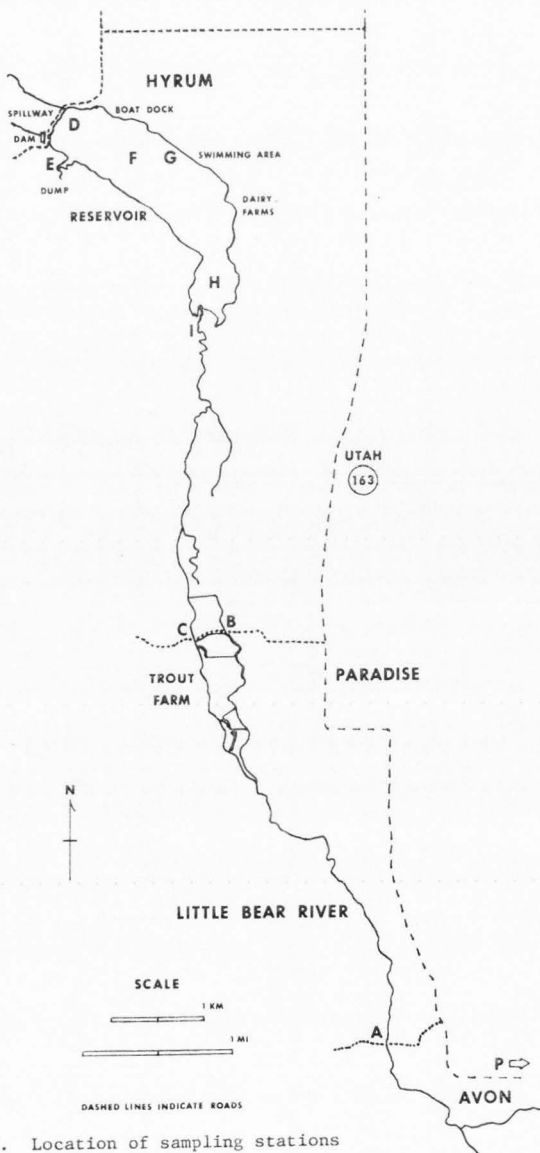


Figure 2. Location of sampling stations

the Hyrum town dump (station E), from the approximate center of the lake (station F), from just off-shore of the swimming area (station G), from the bay area at the southern end of the reservoir (station H), and from the Little Bear River just above the reservoir (station I). No attempt was made to determine or mark the exact locations of stations E through I.

In addition to the above, two locations were sampled once each. A sample was taken from Porcupine Reservoir, located on the east fork of the Little Bear River, upstream from station A. This reservoir is in an unpopulated area, and its waters are considered to be "unpolluted". A sample was also collected from an undesignated spot in the lake (station Bloom) between station D and station F. The sample contained the greatest algal concentration observed during the season.

An inspection of the area surrounding Hyrum Reservoir revealed many possible sources of algal nutrients. On the cliffs along the eastern shore are numerous dairy farms, whose feed-lots overlook the lake, and from which manure could easily be washed into the lake. Cattle were also seen grazing along the western shore. The reservoir is in an agricultural area, and, although not demonstrated, there is a great probability that irrigation run-off finds its way to the lake. Also, as mentioned before, a fish hatchery discharges its effluent into the Little Bear River.

MICROBIOLOGY

From late July until early October, a blue-green alga of the genus Aphanizomenon was the dominant organism of the reservoir. The organism has been tentatively identified as Aphanizomenon flos-aquae (L.) Ralfs. It is the most common of the three species of Aphanizomenon reported in the United States (Prescott, 1970; Reinhard, 1941). Seven species have been described altogether, on the basis of shape and size of vegetative cells, heterocysts, and akinetes. Morphology of the colony has not been used as a species characteristic, as this feature is not constant, varying with conditions. Several strains of Aphanizomenon flos-aquae have also been reported (McLachlan, Hammer, and Gorham, 1963). Prescott (1970) describes colonies of Aphanizomenon flos-aquae as consisting of parallel trichomes, forming a free-floating flakelike bundle, each trichome containing a single heterocyst near the middle and a non-adjacent, intercalary akinete, which appears at maturity.

The bloom has been reported by Lynn (personal communication) to be of such density that when a 16 mm test tube was filled with a water sample, the tube was opaque to transmitted light. During the period of the present study, April to October, 1970, such large concentrations of algae were not observed, but the bloom was quite extensive nonetheless. Associated with the Aphanizomenon were smaller numbers of Anacystis (Microcystis), but these were a minor part of the total algal biomass.

Periodic microscopic examination of water samples showed that in late April large numbers of the diatom Asterionella predominated, followed by numerous other diatoms (Fragilaria, Horomidium, Tabellaria, Cymbella) and the dinoflagellate Ceratium. By the end of May large floating clumps of Lyngbya filaments were common. Some Chlamydomonas were also observed. The algal identifications were made according to Prescott (1970).

The algal population seemed to decline during June, until late July, when the Aphanizomenon became evident in larger and larger numbers. Aphanizomenon concentration fluctuated greatly from time to time and place to place on the lake. The organism appeared to begin to die out in early September but the decline was only temporary. A sudden upsurge in growth resulted in the densest observed Aphanizomenon concentration of the season on September 15th. Carbon analysis of a water sample collected on this date showed 28 mg/l of total organic carbon, much higher than any other sample and about six times the average TOC value. Many other samples contained significant numbers of Aphanizomenon flakes but these were not reflected in the carbon analysis, the amount of carbon in the algae generally being extremely small in relation to the total organic carbon of the water sample.

As the Aphanizomenon grew in the lake they rose to the surface in clumps, and collected along the shoreline and decayed. The decay was evidenced by a light green color and amorphous appearance taken on by the algal clumps. Microscopic examination of the algal mass revealed numerous broken filaments, large amounts of cellular debris, and large numbers of many types of bacteria.

During the bloom period Aphanizomenon flakes were collected for laboratory culture. Collection was made with a plankton net, concentrating the algae in the sample. The samples were carried to the laboratory in an ice chest in "Whirlpak" plastic bags. In the laboratory the flakes were washed by removing them individually with a Pasteur pipet and transferring them to culture media or sterile lake water. They were again transferred in the same way to new medium. The process was repeated as many as ten times, in order to free the Aphanizomenon from other algae and obtain a unialgal culture. No attempt was made to make the algae bacteria-free. As far as is known, this has not yet been accomplished by any investigators.

The algae were cultured in ASM-8a medium (O'Flaherty and Phinney, 1970; composition shown in Appendix); in lake water; in lake water supplemented with nitrate, phosphate, iron, vitamins, or combinations of these; and in lake water autoclaved with lake sediments. The cultures were kept in growth chambers at constant temperatures and under diurnal light cycles. In none of the cultures was significant growth observed, but the algae were maintained for longer periods and in a "healthier" state under certain conditions than under others. The algae survived best in lake water--lake sediment bottles kept at 17 C under low intensity (1500 lux) fluorescent light. Light was supplied for 16 hours per day, but there is no indication that the cycling was of any benefit. Lower temperatures were of definite advantage; cultures at temperatures over 20 C lysed rapidly, but this may have resulted from contaminating bacteria growing more rapidly at

the higher temperatures rather than being due to any harmful effect of the higher temperatures on the algae. (The lake temperature during the bloom period was observed to vary between 18 C and 24 C.)

Maintenance in filtered lake water was better than in ASM-8a medium. Adding nutrients or ASM-8a medium to the lake water decreased survival time in direct proportion to the amount of nutrient or medium added. Bubbling air through the cultures appeared to be neither harmful nor beneficial. Nor did making transfers appear to be of any value, the transferred material often lysing before the parent culture.

O'Flaherty and Phinney (1970) reported success in growing Aphanizomenon flos-aquae and maintaining it in the natural flake form for more than three years, using ASM-8a medium, a modification of the ASM medium of McLachlan and Gorham (1961). McLachlan, Hammer, and Gorham (1963) were able to maintain the flake form for several months in ASM with soil extract, but after a prolonged period the alga became partially or completely non-colonial. Ten strains of Aphanizomenon flos-aquae were tested, and growth patterns varied with strain. Gentile and Maloney (1969) reported different pH optima for different strains of Aphanizomenon. In the present study pH was not tested as a variable.

There have been frequent reports of waterblooms of Aphanizomenon in the United States, often in lakes receiving runoff from agricultural areas (McLachlan, Hammer, and Gorham, 1963), as does Hyrum Reservoir. The above authors suggested that iron leached from soils may be responsible for large Aphanizomenon colonies. Gentile and Maloney (1969) reported that Aphanizomenon as well as other blue-green algae

are capable of producing blooms at very low phosphorus concentrations, 10 $\mu\text{g P/l}$ being the critical level of that element for the development of nuisance growths. They also cited evidence that Aphanizomenon is capable of nitrogen fixation.

Recently, a potent toxin has been reported to be released from natural populations of Aphanizomenon when the cells lysed (Sawyer, Gentile, and Sasner, 1968; Gentile and Maloney, 1969). The toxin is a nerve and muscle-blocking agent which destroys conduction in these tissues, and has definitely been shown to be toxic to fish under experimental conditions. It is possible that such toxins may prove a hazard to swimmers during bloom periods (Schwimmer and Schwimmer, 1955).

DISSOLVED OXYGEN AND TEMPERATURE

Dissolved oxygen (DO) and temperature profiles were taken at station D on six days during the course of the study. The measurements were obtained using an Electronic Instruments Limited (E.I.L.) model 15A dissolved oxygen meter with a model 15A biological oxygen electrode and a thermistor on a 24 meter cable. Readings were taken in percent saturation (temperature compensated), and these values were used to calculate the dissolved oxygen concentration in mg/l. Saturation values for Cache Valley, Utah (elevation 4,700 feet, mean barometric pressure 0.85 atmospheres) were calculated from the sea-level values in Hutchinson (1957) by correcting by a factor of 0.85, and are presented in the Appendix.

The instrument was calibrated by placing the electrode in water being sparged with air, and adjusting the meter reading to 100% saturation. The accuracy of the instrument was checked by comparing its readings with Winkler tests (APHA, 1971) performed on the same samples. No discrepancies were noted. Thermistor calibration was also verified in the laboratory.

The profiles were taken by lowering the electrodes into the lake and noting the meter readings at one-half to three meter intervals, depending on the rates of change. Readings were again made as the electrodes were drawn up from the bottom. If a discrepancy was observed between the two sets of readings, the process was repeated.

The June 14 profile (Figure 3) shows a gradual decrease in oxygen concentration with depth, with a value of 5.9 mg/l at the bottom

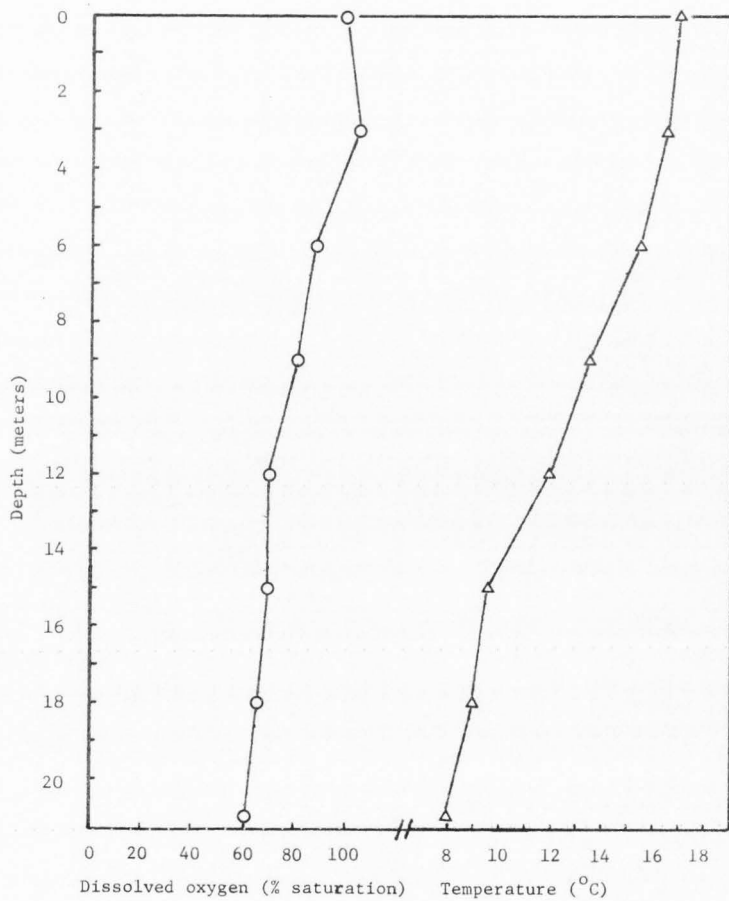


Figure 3. Dissolved oxygen and temperature profiles, station D, June 14, 1970.

(21 meters). The second profile, taken August 13, (Figure 4), shows supersaturation, (9.0 mg/l) down to 5 meters, with a sudden decrease to 3.5 mg/l at 5.5 meters. The decrease continued more gradually below this depth, reaching anoxic conditions at 14 meters. The August 18 and 28 profiles (Figures 5 and 6) are similar to August 13, except that the sudden decrease began at 6 meters. By September 11 (Figure 7) the decrease was much less pronounced and began at 13 meters. The bottom of the lake had also become oxygenated again. The October 10 profile (Figure 8) straightened even more, and appeared to be approaching conditions encountered in the first profile on June 14.

These same profiles are shown in three-dimensions with oxygen concentration, depth, and time as coordinates, in Figure 9. The change in conditions with time, and the apparent seasonal nature of the phenomenon are evident. Oxygen depletion occurred in the hypolimnion during the summer months, returning to more oxygenated conditions in September. Some measurements by Porcella (personal communication) indicated that some mixing and oxygenation of the hypolimnion did occur during a storm. It is not known how quickly anoxic conditions were reestablished.

The most unusual feature of these measurements is the extremely sharp drop in oxygen concentration exhibited in mid-summer. On August 13 (Figure 4) at 5 meters depth the water was 115% saturated with oxygen; at 5.5 meters the value was 48%. This was not accompanied by any measured change in temperature. At no time was any large thermocline observed, the most pronounced being recorded on August 18, (Figure 5) when the temperature dropped from 21.5 C to 20 C over a three meter span, or one-half degree C per meter.

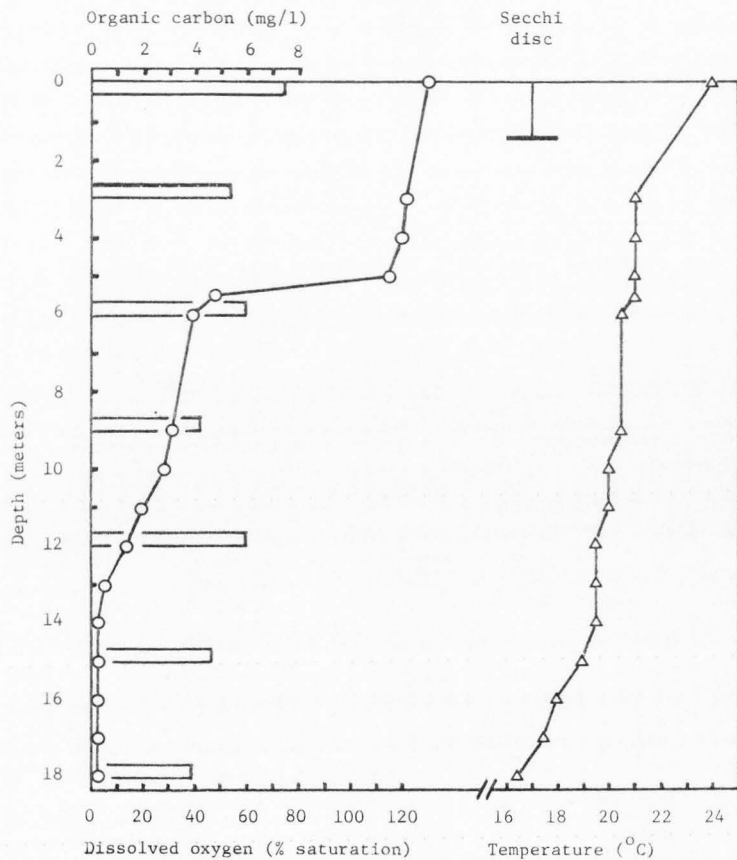


Figure 4. Dissolved oxygen, temperature, and organic carbon profiles, and Secchi disc transparency, station D, August 13, 1970.

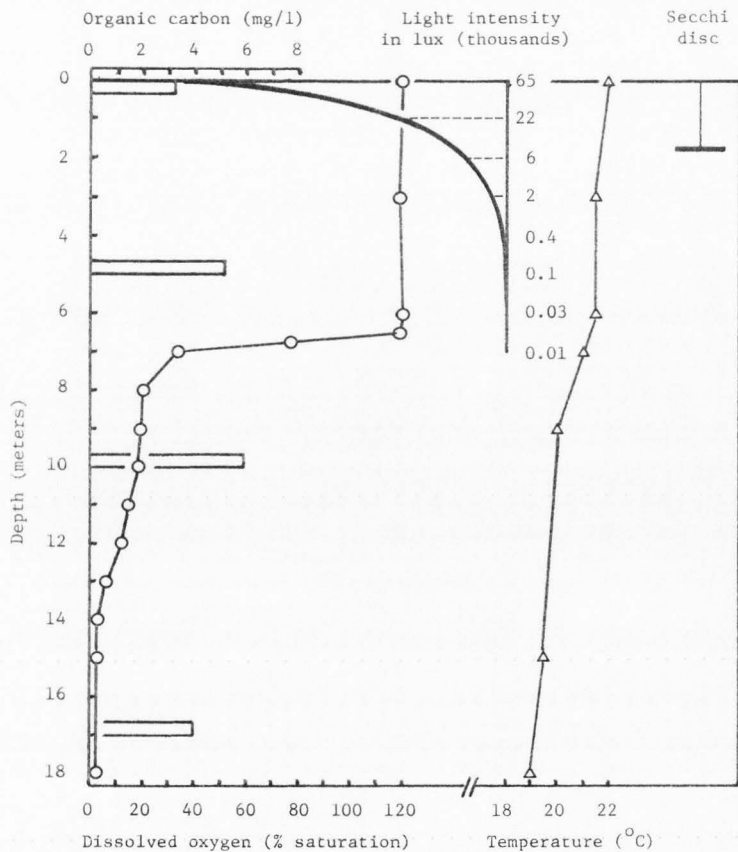


Figure 5. Dissolved oxygen, temperature, organic carbon, and light intensity profiles, and Secchi disc transparency, station D, August 18, 1970.

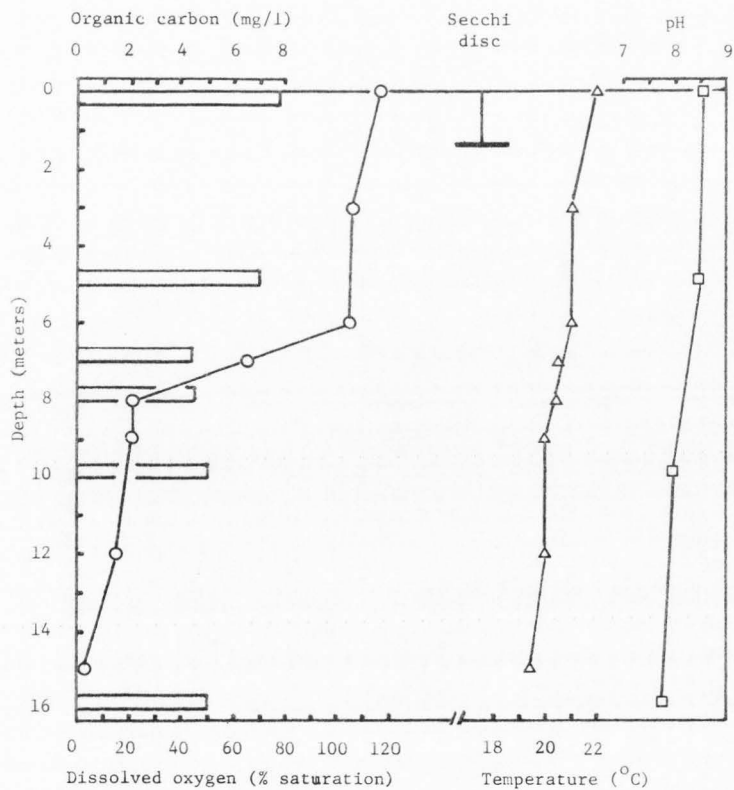


Figure 6. Dissolved oxygen, temperature, organic carbon, and pH profiles, and Secchi disc transparency, station D, August 28, 1970.

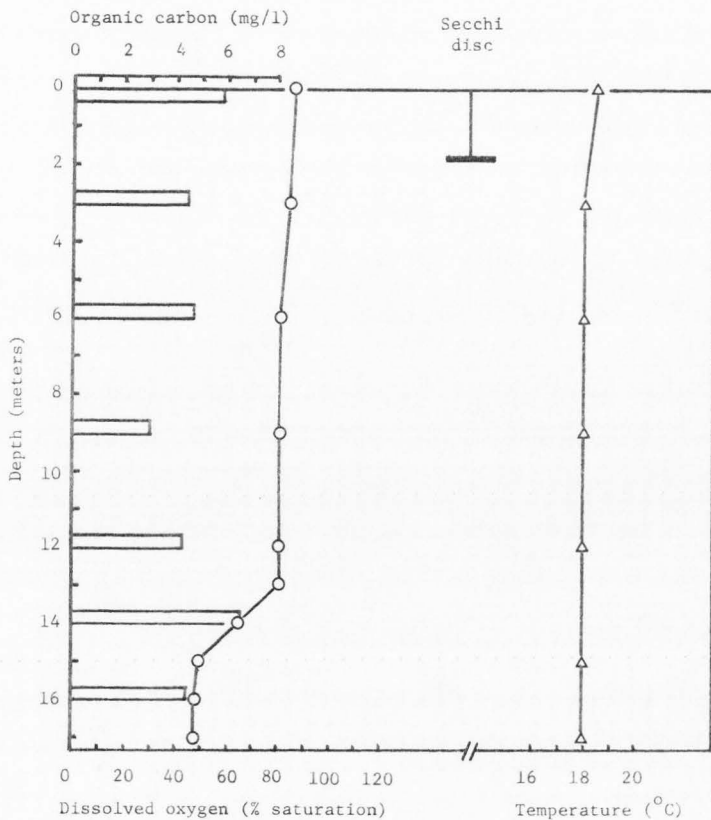


Figure 7. Dissolved oxygen, temperature, and organic carbon profiles, and Secchi disc transparency, station D, September 11, 1970.

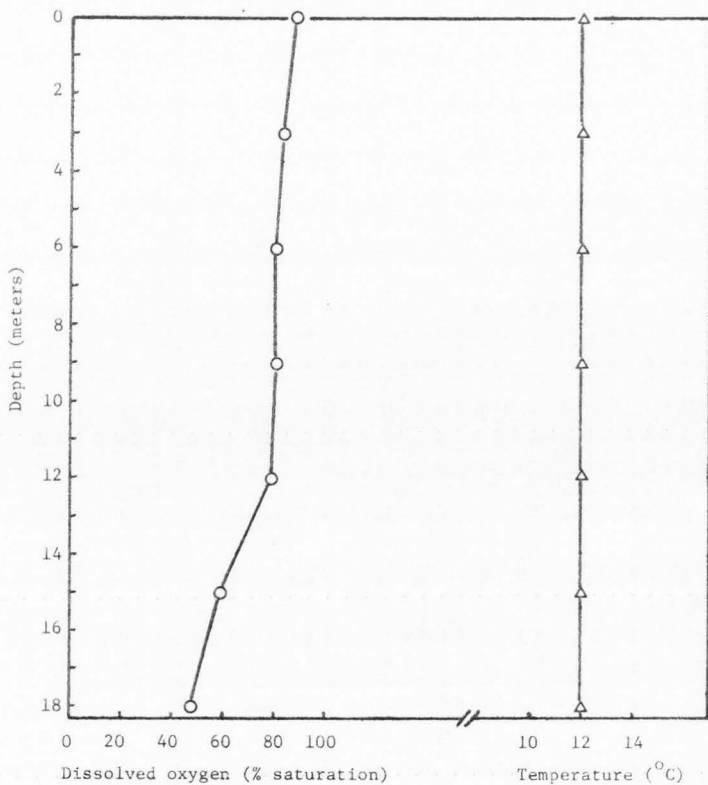


Figure 8. Dissolved oxygen and temperature profiles, station D, October 10, 1970.

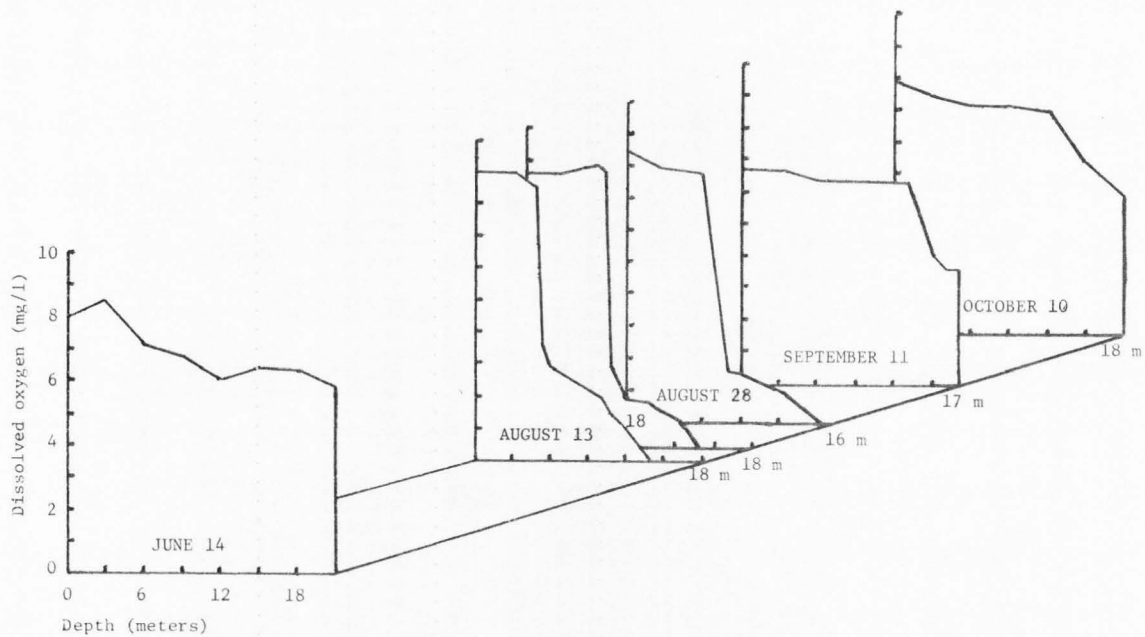


Figure 9. Dissolved oxygen versus depth and time, station D.

The stability of the oxycline in the absence of a strong thermocline is explained by the relative thermal resistance of water to mixing. At 20 C twenty times as much energy is required to mix two masses of water with a temperature differential of one degree C than would be required at 5 C. A barely perceptible temperature difference (0.1° C) in warm water results in a great resistance to mixing (Valentyne, 1957). The low dissolved oxygen levels below the oxycline are undoubtedly the result of biological activity, causing a depletion of the available oxygen, and the supersaturation above the oxycline could be attributed to algal photosynthesis. But the extreme sharpness of the boundary between the two regions appears to be unusual. Perhaps the use of an in situ probe has revealed a phenomenon that is not ordinarily made evident by conventional discrete sampling procedures.

CARBON

One hundred fifty-one water samples were collected from eleven locations on twenty days (Table 2) during the period of the study. Thirteen of the samples were collected in duplicate. The sampling times were between 1100 and 1500 hours. All stations were sampled at the surface and additional samples (Table 2) were taken at varying depths at station D, the deepest part of the reservoir, at the same time that the dissolved oxygen, temperature, and other readings were made.

Surface samples were collected directly in 6 or 18 ounce "Whirlpak" plastic bags. Depth samples were taken in a one-liter brass Kemmerer type sampler and transferred to "Whirlpaks". The samples were kept cool in an ice chest for one to four hours until they could be returned to the laboratory where they were frozen. The frozen state was maintained until the samples were thawed for analysis, four to eleven months later, depending upon the dates of collection and analysis. Storage temperature varied from -18 C to 2 C. Due to malfunction of the freezing unit, the stored samples partially thawed for a brief period, and some material was lost, but the organic carbon values did not appear to be affected. A control to determine the effects of freezing and storage in plastic containers on the organic carbon in the water samples produced equivocal results. There was some indication of a slight loss in organic carbon, probably through adsorption to the container walls.

Table 2. Samples collected

Date	Location
4/24/70	A, B, C, D.
5/1	A, B, C, D.
5/8	A', B', C', D'.
5/14	A, B, C, D.
5/17	D, E, F, G, H, I.
5/24	A, B, C, D, E, F, G, H.
6/7	A, B, C, D, E, F, G, H.
6/14	A, B, C, D, E, F, G, H.
7/30	A, B, C, D, E, F, G, H.
8/13	A, B, C, D-s, 3, 6, 9, 12, 15, 18, E, F, G, H.
8/18	D-s, 5, 10, 17.
8/28	D-s, 5, 7, 8, 10, 16.
8/30	A', B', C, P'.
9/9	D, E, F', G, H'.
9/11	D-s, 1, 3, 6, 9, 12, 14, 16.
9/15	Bloom
9/21	A, B, C, D.
10/3	A', B', C', D'.
10/10	D, E, F.
10/18	A, B, C, D'.

All samples are sub-surface "s" unless otherwise indicated by numerals showing depth, in meters. ' means sample was collected in duplicate.

To prepare for analysis, the frozen sample was placed in an acid washed glass beaker in an oven at 80 C until a small amount of ice remained. The still cool sample was then stirred with a teflon coated stirring bar on a magnetic stirrer, and a small quantity of the mixed sample was poured into an acid washed glass collecting vial. The material was then homogenized in the collecting vial by subjecting it to four 10-second bursts from a Bronwill biosonicator, at maximum power for the tip used. A glass encased magnetic stirring bar was placed in the vial and the material was stirred on a magnetic stirrer as samples were withdrawn for analysis. The samples were not filtered for separate determinations of dissolved and particulate organic content, because the freezing and thawing process had disrupted the particulate material causing a release of cellular material into solution.

The analysis was performed on a Beckman model 915 total organic carbon analyzer, according to the method of Van Hall and Stenger (1967). Twenty μ l aliquots were taken from the collecting vial with a 50 μ l Hamilton syringe and injected into either the total carbon or inorganic carbon channel of the carbon analyzer. Results were read directly in milligrams of carbon per liter (mg C/l) on the recorder; the readings of three to five replicate injections of each sample were averaged, and the values obtained were used to calculate the total organic carbon (TOC) concentration by subtracting the inorganic carbon value from the total carbon reading. The repeatability of analyses by the instrument was within a range of 2% of full scale, or 2 mg C/l. This conclusion

was supported both by the instruction manual (Beckman, 1968) and by daily injection of a standard, which showed a range of 1.9 mg TOC/l.

A total of 118 total organic carbon values were obtained. The mean value was 4.62 mg/l, with a range of from 1.2 to 8.9 mg/l, (Figure 10). The standard deviation (s) was 1.47 and the population mean was calculated, at 95% probability, to lie between 4.36 and 4.88 mg/l. Statistical analyses were done after Sokal and Rohlf (1969).

The data were examined for trends and correlations. First they were grouped into samples from three periods: 4/24 to 5/24, 8/13 to 8/30, and 9/9 to 10/18 (Figure 11). Means and standard deviations for each of the groups were calculated, and the means were compared for differences. The t-tests between the means (95% confidence limits) showed that the differences were not significant, i.e., TOC did not vary with season. Comparison was also made between means of samples from stations A, B, C, and D (Figure 12). There were shown to be no significant differences in organic carbon values between these stations.

Regression analyses were made of the correlations between total organic carbon concentration and depth (Figure 13), and between TOC and dissolved oxygen (Figure 14). TOC showed a slight decrease with depth (correlation coefficient -0.50), but no correlation between TOC and DO was observed. The total organic carbon values are shown in conjunction with the concurrent dissolved oxygen and temperature profiles in Figures 4-7.

Plots of TOC fluctuation with time at stations A, B, C, and D are shown in Figure 15. At stations A and C, along the Little Bear River, there was an apparent decrease in TOC as the season progressed;

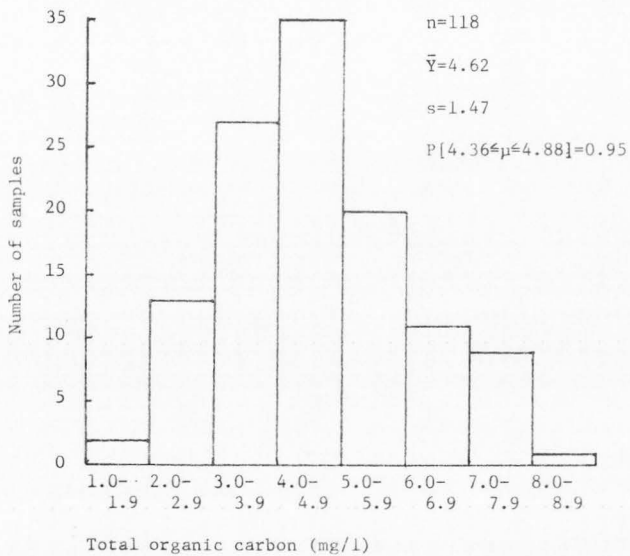


Figure 10. Total organic carbon analysis: all samples.

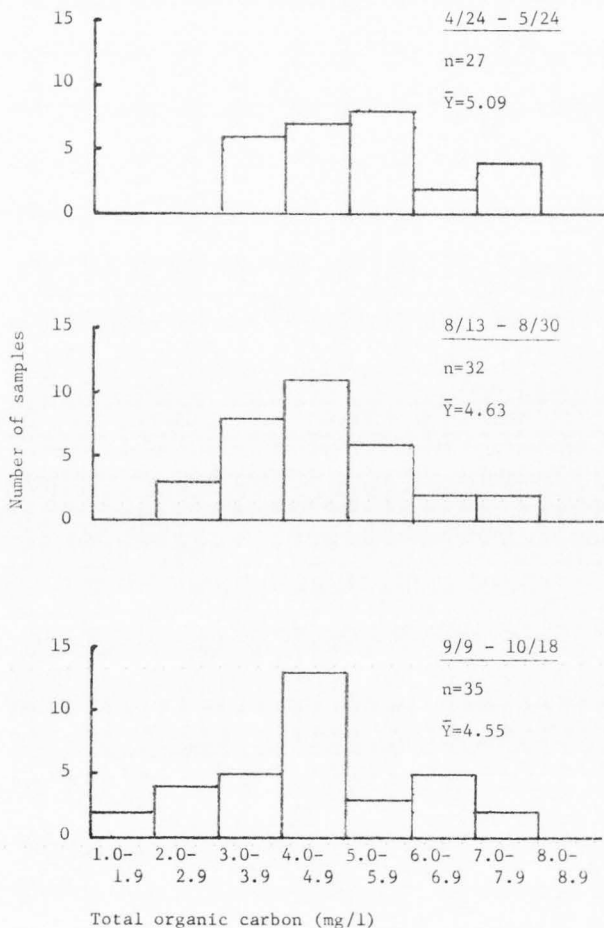


Figure 11. Total organic carbon analysis: spring, summer, and fall periods.

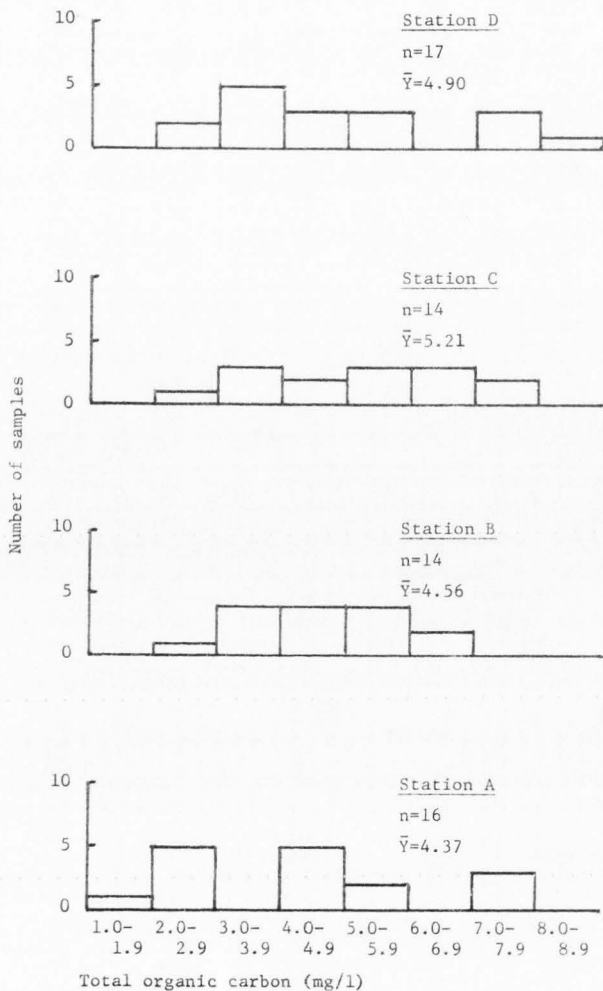


Figure 12. Total organic carbon analysis: stations A,B,C, and D.

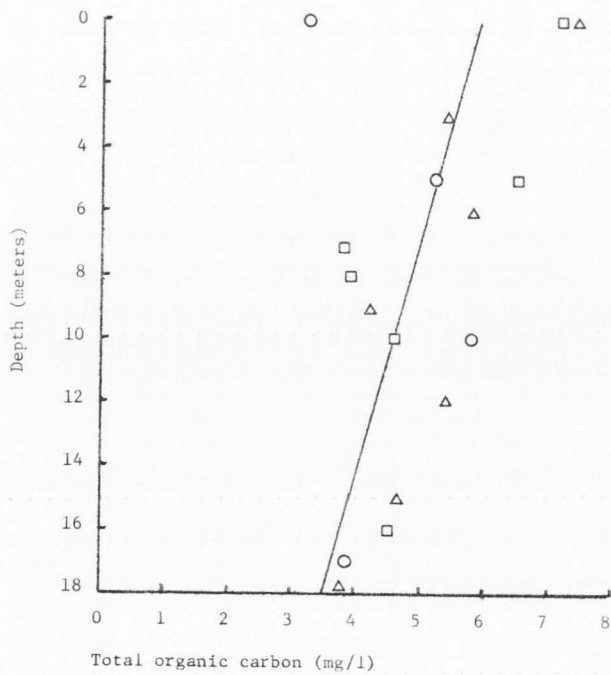
Δ 8/13 \circ 8/18 \square 8/28 $r = -0.50$ 

Figure 13. Total organic carbon versus depth: station D.

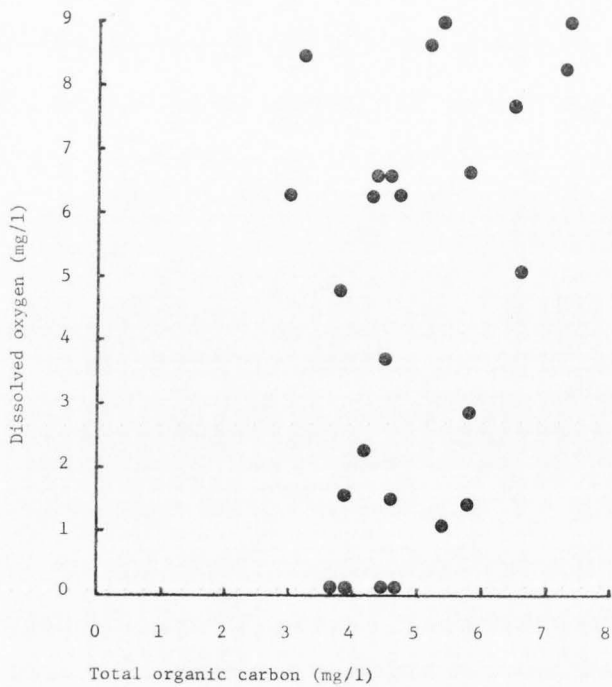


Figure 14. Total organic carbon versus dissolved oxygen.

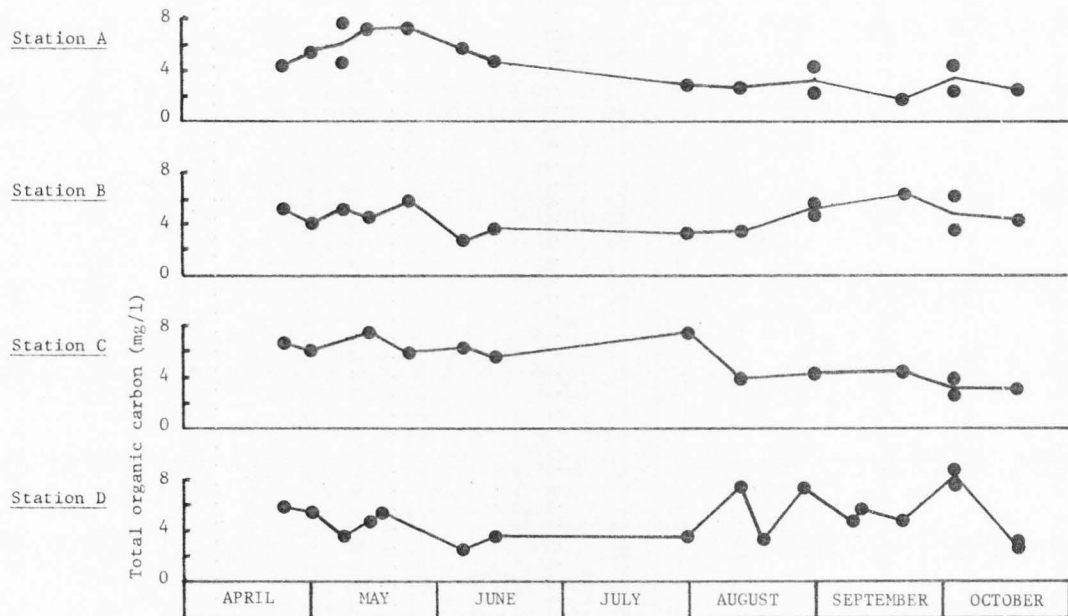


Figure 15. Total organic carbon: stations A, B, C, and D versus time.

at station B, the outlet from White's Trout Farm, the value remained constant; and at station D, the deepest part of the reservoir, TOC appeared to increase slightly during the bloom period from late July to early October. However, each point on these graphs represents only a single sample and, because the repeatability of the instrument is only within a 2 mg C/l range, it would seem prudent not to draw any conclusions from these graphs before repeating the study with concentrated samples. A similar situation holds for Figure 16, which shows TOC at the various stations for different dates. Some trends appear; early in the season TOC increased from station A to station C, and then decreased again at station D, whereas during the bloom period station D had more organic carbon than the first two stations. The intermediate stations did not exhibit any regularity of change. The peak in TOC along the Little Bear River in the spring correlated directly with stream discharge (USDI, 1971); but the correlation was considerably better at station A (Figure 17) than at station C. As the stream flow decreased during the summer, so did the TOC concentration. High stream velocities churn up sediments which are rich in organics and contribute to the TOC (Leopold and Maddock, 1953).

In summation, the total organic carbon concentration in Hyrum Reservoir and its tributary, the Little Bear River, remained virtually constant during the study period. There was a decrease in TOC concentration in the Little Bear River as the season progressed, due to decrease in stream flow. Slight changes in TOC concentration in the reservoir itself were not confirmed, namely (1) an increase in late summer, probably attributable to the increase in algal biomass; and

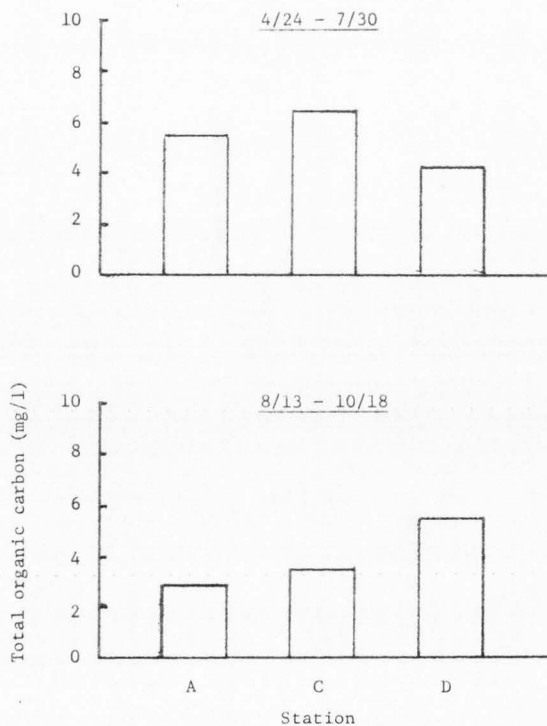


Figure 16. Total organic carbon fluctuation with station: early versus late summer.

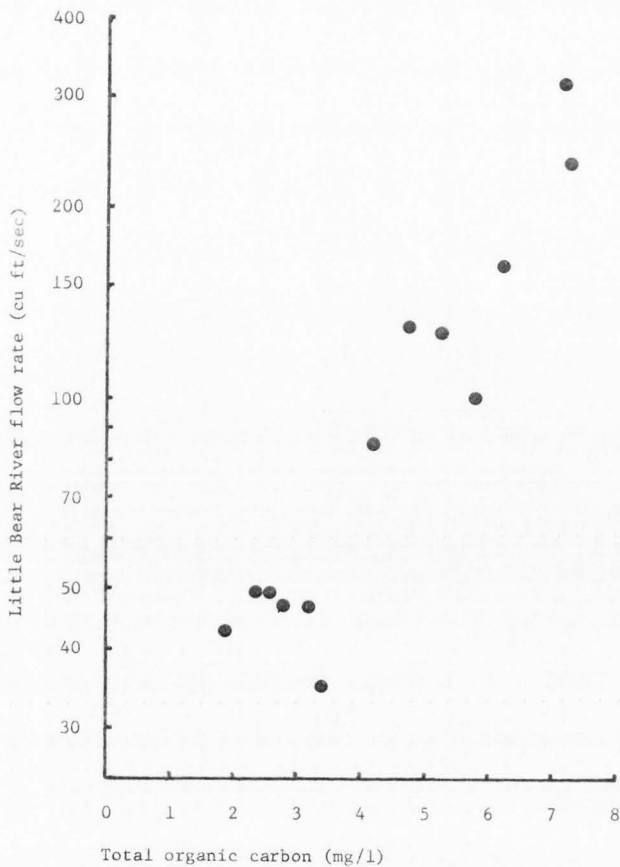


Figure 17. Total organic carbon versus stream discharge at station A.

(2) a decrease with depth, which may have been the result of biological oxidation of organic matter. There was no correlation between TOC and dissolved oxygen, which decreased with depth.

The increase in organic carbon at station D during the late summer would appear to be linked to the algal bloom, but no independent measurements of algal biomass are available to corroborate this. Three samples, 5/24 D, 5/24 E, and 9/15 Bloom contained large masses of algae, and these gave extremely high TOC values (Table 3); however, other samples also contained significant numbers of Aphanizomenon flakes without showing significantly increased organic carbon values. Apparently, the algae contributed very little to the total organic carbon concentration in these waters unless the algal biomass was exceptionally high, approaching that of a dense laboratory culture. Even during the bloom, unless the sample was taken from an area with a dense patch of algae, the algal organic carbon was a relatively small percentage of the total organic carbon in the water.

There have not been a great many studies of organic carbon in natural waters, but the results that are available all tend to corroborate the conclusions of the present investigation. Birge and Juday (1927) analyzed the water of 84 Wisconsin lakes for organic matter and found a mean value of 7.3 mg TOC/l with a range of 2.0 to 20.9 mg TOC/l. Of this, the dissolved organic carbon averaged 6.4 mg/l with a range of 1.4 to 19.8 mg/l, the balance being particulate. Dissolved organic carbon was defined by Birge and Juday as that which remained after centrifugation, and the authors claimed that centrifugation removed 95% of the particulate matter larger than bacteria, plus 25 to 50% of

Table 3. Carbon analysis of samples containing large algal masses.

Date	Station	Depth	Total organic carbon (mg/l)	Alga present
5/24	D	surface	21.8	<u>Lyngbya</u>
5/24	E	"	13.3	"
9/15	Bloom	"	28.4	<u>Aphanizomenon</u>

the bacteria. The water was then evaporated and the residue weighed. The percentage of organic matter in the residue ranged from 20 to 80%, and 50% of this organic matter was assumed to be carbon.

A second study by Birge and Juday (1934) included 529 lakes, and reported a mean value of 7.65 mg TOC/l, with a range of 1.15 to 28.5 mg TOC/l. The planktonic (particulate or centrifugable) carbon represented from 3 to 24% of the total organic carbon. Birge and Juday concluded that dissolved organic matter is a fairly definite quantity for a particular lake, not showing great variation either with depth or time. Particulate matter was found to comprise a relatively small percentage of the total organic content, and the percentage decreased as TOC increased. Where dissolved organic carbon was 4-6 mg/l, it was about six times the weight of the plankton. A large, temporary crop of algae raised the percentage of plankton in the total.

Birge and Juday (1927) likened the organic matter situation in a lake to that in the soil, where the humic material is fairly constant in quantity and composition, is not readily decomposed beyond a certain point, and persists with little change for a long time. Superimposed upon this is the field crop, analogous to the plankton, with its small quantitative relationship to the total organic matter. Therefore, they suggested that because the dissolved organic carbon remains fairly constant, a single carbon analysis is sufficient to put a lake in its true place in its organic relations, and to classify it.

Riley (1940), in studies on particulate organic carbon in Linsley Pond, Connecticut, found a range of 0.6 to 2.1 mg C/l, with

a mean of 1.2 mg C/l. The variations in particulate organic matter were found to be smaller than the variations in chlorophyll or phytoplankton volume, but the seasonal cycles were similar. He concluded that phytoplankton accounted for the major variations in the weight of particulate organic matter, and that the total quantity of other constituents -- zooplankton, bacteria, and detritus -- was relatively constant. Dissolved organic matter was not reported.

Forsberg (1967) measured the dissolved organic carbon in some lakes in Uppland, Sweden, using the technique of Menzel and Vaccaro (1964). In this method the organic carbon in a sample is oxidized with potassium persulfate in sealed glass ampoules, and the CO_2 released is measured with an infrared analyzer. The organic carbon concentrations found in glass-fiber filtered lake samples ranged from 5 to 17 mg/l, with variations of around 2 mg/l in any particular lake. No correlation was found between dissolved organic carbon and dissolved oxygen, total phosphorus, or total nitrogen. A negative correlation was observed between chlorophyll (a + b) and dissolved organic carbon, which dropped slightly during an algal bloom. No examination or mention was made of the particulate carbon.

Weber and Moore (1967) studied particulate organic matter and dissolved organic carbon in a small, hardwater, midwestern stream. Particulate matter was removed from samples by centrifugation, then dried, ignited, and weighed, the weight loss being a measure of the organic content. A correlation was found between particulate organic matter and stream discharge during the period from December to April, and a negative correlation was observed from May to November. This

was interpreted to mean that the particulate organic matter was not derived from the same sources during the two seasons, the positive correlation showing that the organic material was scoured from the river bed, and the negative correlation indicating that the organic matter during the summer months was autochthonous and probably of phytoplanktonic origin. Dissolved organic carbon was determined by passing samples through a 0.45 μ pore diameter Millipore filter, acidifying the filtrate to pH 2, and analyzing with a "Beckman carbonaceous analyzer". The concentrations of dissolved organic carbon ranged from 2.5 to 12.5 mg/l and averaged 6.4 mg/l. No seasonal patterns were apparent, and a low correlation, (-0.061), was found between dissolved organic carbon and phytoplankton volume during the season when the standing crop of algae was high, indicating that the phytoplankton was not a major source of dissolved organic matter. Nor was any relationship found between dissolved organic matter and river discharge.

A comparison of the distribution of organic matter in the five Great Lakes by Robertson and Powers (1967) showed the amounts of dissolved organic matter to be three to ten times larger than the particulate organic matter, which in turn is much greater than the amounts of zooplankton and macrobenthos. In Lake Superior the dissolved organic carbon was 1.1 to 1.5 mg/l and the particulate organic carbon was 0.1 to 0.3 mg/l. The values for Lake Ontario were 2.9 to 3.3 mg/l for dissolved organic carbon and 0.5 to 0.8 mg/l for particulate organic carbon. The other lakes ranged between these

values. (The results were reported in terms of organic matter, and have here been converted to organic carbon by multiplying by a factor of 0.5.)

Brooks (1970), using the method of Menzel and Vaccaro (1964), measured the distribution of organic carbon in the Brazos River basin in Texas. He found that the dissolved organic carbon (DOC) concentration ranged from 2.8 mg/l to 7.0 mg/l, and the particulate organic carbon (POC) concentration ranged from 1.0 mg/l to 16 mg/l. The DOC concentrations were found to be more independent of flow rate than the POC concentrations, which were directly related to river discharge. He concluded that DOC values were the best indication of organic water pollution, as the POC from domestic pollution was generally broken down before it reached the river, while the DOC was more resistant to degradation.

Most of the recent work on dissolved and particulate organic carbon has been done in the marine habitat. Menzel (1967), concluded from his studies that (1) surface standing crop has no measurable influence on the concentration of organic particles occurring at depth; (2) below a given depth the distribution of these particles is homogeneous in time, space, and depth; and, (3) there is no consistent decrease in dissolved organic carbon with depth. But Duursma (1965) reports that there is generally more organic matter in the surface layers than in the deeper water, and that distribution in the upper layers depends somewhat on season, and probably also on water movements.

Parsons (1963) expressed on a relative scale based on 100 the approximate distribution of the total organic matter in the sea as

follows: soluble organic 100, particulate detritus 10, phytoplankton 2, zooplankton 0.2, and fish 0.002. Mullin (1965) found less than 10 to 20% of the total carbon at any station to be in the form of living phytoplankton, and Strickland (1965) reported that the quantity of dissolved organic matter passing through a 0.5 μ pore diameter membrane filter nearly always exceeded the amount of particulate organic material by a factor of 10 or more, and 50 or more if only living cells were considered.

Riley (1963) described the nonliving, particulate organic matter in seawater as consisting of delicate, platelike aggregates ranging in size from about 5 μ to several millimeters in diameter, the aggregates being amorphous matrices containing both organic and inorganic materials, with inclusions of bacteria and phytoplankton. According to Riley (1963) and others (Baylor and Sutcliffe 1963; Riley, Wangersky, and Van Hemert, 1964; Sutcliffe, Baylor, and Menzel, 1963) the aggregates are formed mainly by adsorption of dissolved organic matter on bubbles and on other naturally occurring surfaces. The aggregates are presumed to act as a substrate for bacterial growth and to provide food for larger organisms.

The dissolved organic matter in lakes is usually derived from the phytoplankton which have developed in the lake and which can be the source of up to six times their own weight of dissolved organic materials, through excretion and decay (Birge and Juday, 1934; Fogg, 1962; Hellebust, 1965; Strickland, 1965; Kuznetsov, 1968). Additional organic material, generally highly colored, is often extracted from peat and soils in the catchment area of the lake and carried to the

lake by streams and runoff (Hutchinson, 1957). One could expect to find almost any organic compounds in the dissolved organic matter, including carbohydrates, fatty acids, amino acids and other nitrogen containing organics, enzymes, vitamins, auxins, antibiotics, and toxins (Provasoli, 1963).

These compounds undoubtedly play a large role in the ecology of the lake. Although the ability of algae to grow heterotrophically below the photic zone has been disputed (Provasoli, 1963; Wright and Hobbie, 1966), the need for organic growth factors by many algae has been more conclusively demonstrated (reviewed by Provasoli, 1963). However, the low concentrations of dissolved organic compounds found in natural waters are more than sufficient to promote the growth of heterotrophic bacteria (ZoBell and Grant, 1943; Wright and Hobbie, 1966), which also fix carbon from free carbonic acid during heterotrophic growth (Kuznetsov, 1968). Fogg (1962) has suggested that organic nitrogen compounds manufactured and excreted by blue-green algae are utilized by other organisms associated with the alga. In addition, he maintained that organic acids and polypeptides excreted by algae chelate inorganic ions, thus maintaining nutrient substances such as iron and phosphate in solution, making them more readily available for algal growth. The chelation of copper by these organic substances would reduce its toxic effect by reducing its effective concentration, thus explaining the lack of effectiveness of some copper sulfate treatments in eliminating algal nuisance situations.

OTHER MEASUREMENTS

Several measurements on the reservoir were taken in addition to organic carbon and dissolved oxygen. Secchi disc readings were taken in conjunction with four of the oxygen profiles (Figures 4-7). The Secchi disc depths ranged from 1.3 to 1.8 meters. On August 18 a submarine photometer (Fred Schueler Co.) was used to determine the depth of the photic zone, and the data obtained is reproduced in Figure 5. The incident light at the surface was 65,000 lux; it decreased logarithmically to 1% of this value at 3.5 meters; and at 7 meters 10 lux were recorded. At the Secchi disc depth of 1.7 meters the light intensity was approximately 9,000 lux, or 14% of the incident light. This is in agreement with the work of Beeton (1958), who found the average percentage transmission of surface light intensity, at the Secchi disc depth, to be 14.7%.

On August 28, pH readings were made on lake-water samples taken from the surface and depths of 5, 10, and 16 meters. The pH decreased slightly from 8.6 at the surface to 8.5 at 5 meters, then dropped to 8.0 at 10 meters, and to 7.8 at the bottom (Figure 6). The higher pH in the trophogenic zone is due to the uptake of CO_2 by phytoplankton, and the reverse process occurs in the tropholytic zone of the hypolimnion where CO_2 liberated by microorganisms causes a lowering of pH. The degree of pH lowering is controlled by the bicarbonate alkalinity of the water. "When the bicarbonate alkalinity is high and the trophogenic zone productive, the consequent high production

of CO_2 in the hypolimnion causes a relatively small lowering of the pH of the well-buffered water.", (Hutchinson, 1957, p. 685).

PROPOSED ADDITIONAL STUDIES

The present investigation has revealed some interesting facets of the microbiology and limnology of Hyrum Reservoir which could profitably be studied further. The sharp oxycline should be investigated in detail; its shape should be recorded weekly throughout the year, and several sets of diurnal readings should also be taken, in order to better define the fluctuations which occur, and to determine if any mixing occurs during the summer stratification. pH values should be recorded immediately above and below the oxycline, and an attempt should be made to describe and enumerate the bacterial populations associated with, and occurring just above and below, the oxycline.

The determination of the culture conditions necessary for maintenance of the Aphanizomenon in the laboratory has been the first step in the process which should include isolation of the alga in a unialgal culture, and growth in a defined artificial medium, perhaps a modification of ASM-8a (Appendix). The ultimate goal, of course, is to determine the limiting nutrient(s) in the lake water. This would probably best be done by using bioassay techniques (EPA, 1971).

It has been shown in the present investigation that the Beckman model 915 total organic carbon analyzer is not sufficiently sensitive for direct measurement of organic carbon concentrations in lake waters. However, by use of suitable sample concentration methods it should be possible to obtain meaningful organic carbon values with this

instrument. A study of the organic carbon relationships in Hyrum Reservoir would certainly be of great interest and value, and it is suggested that a suitable technique be developed and applied. This is an important research area which is only beginning to receive attention. For a review of recent work, see Hood (1970).

SUMMARY AND CONCLUSIONS

A study was undertaken to determine the causes of a dense late-summer waterbloom of Aphanizomenon flos-aquae in Hyrum Reservoir, Cache Valley, northern Utah, and to suggest some possible corrective measures. A sampling program was established and samples were taken from late April 1970 until early October of that year at several stations in the reservoir and along its tributary, Little Bear River. Samples were also taken of the effluent from White's Trout Farm, on the Little Bear River, a suspected source of algal nutrients. Surface samples were collected at all stations; in addition, depth samples were taken at station D, the deepest part of the reservoir, near the outflow.

The samples were stored frozen, then thawed and analyzed for total organic carbon (TOC) concentration on a Beckman model 915 total organic carbon analyzer. It was found that TOC concentration did not vary significantly with time or place of sampling. There was a correlation between TOC and stream discharge along the Little Bear River, due to the increased sediment load at high discharge rates; and a slight decrease in TOC with depth at station D was also observed. TOC also appeared to increase slightly at station D during the summer months, but the increase was not based on sufficient samples to establish its statistical significance. The mean value for all samples (n=118) analyzed was 4.62 mg TOC/l, with a range of from 1.2 to 8.9 mg TOC/l and a standard deviation of 1.47.

The algal bloom was not reflected by the TOC measurements. One sample, collected from a thick patch of algae during the height of the bloom (September 15) gave a TOC value of 28.4 mg/l, but other samples containing smaller numbers of algae did not differ significantly from the overall mean value. A literature review revealed that other investigators have encountered similar situations, and the consensus of these investigators is that in natural waters the particulate organic matter constitutes only a small fraction (10-30%) of the total organic matter in the water. There exists a residuum of dissolved and colloidal organic material which remains relatively constant in concentration from place to place and time to time, and which is much greater in magnitude than the standing algal crop. The dissolved material is presumed to play a significant ecological role in maintaining the balance of the system in the face of the large fluctuations in individual species that occur periodically.

Although it was found that total organic carbon is not a good parameter for monitoring changes in the system, the TOC value does give an indication of the quality of the lake in relation to other lakes. A fixed value such as this would be useful in comparing lakes if analytical methods were standardized and organic carbon data from numerous lakes made available, along with other information about the lakes.

All of the information gathered in the present study supports the conclusion that Hyrum Reservoir is highly eutrophic. Hyrum has a higher total organic carbon concentration (4.6 mg/l) than Lake Erie (4.0 mg/l) and Lake Ontario (3.8 mg/l), the most eutrophic of the

Great Lakes. The oxygen supersaturation in the trophogenic zone coupled with its depletion in the hypolimnion is another indication of high productivity and high biological activity. This is also confirmed by the low light penetration and the pH changes observed.

The greater problem of determining the specific cause of the Aphanizomenon bloom remains; and some techniques for accomplishing this have been outlined in the preceding section. Finding and eliminating a limiting nutrient may solve the problem, or it may allow another alga to replace the present nuisance organism. A simpler solution might be the use of copper sulfate or some other chemical agent, but this also might prove to be ineffective. In addition, it could result in some unwanted side-effect, such as a sudden release of algal toxin, causing fish mortalities or bathers' rash.

Another approach is to stop the flow of algal nutrients into the lake. This would entail channeling all agricultural runoff directly to the outflow of the reservoir, removing the cattle from the shore line, and possibly treating the effluent from the trout farm to remove nutrients. This approach, although expensive, might be the most satisfactory in the long run if the reservoir is to be maintained as a recreational area.

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APPENDIX

Table 4. Composition of ASM-8a Aphanizomenon medium

Compound	Molecular weight	Milligrams per liter	Micromoles per liter
<u>Major elements</u>			
MgSO ₄ ·7H ₂ O	246.5	50.0	200
CaCl ₂ ·2H ₂ O	129.0	15.0	100
NaNO ₃	85.0	85.0	1000
NaHCO ₃	84.0	10.0	120
MgCl ₂ ·6H ₂ O	203.3	80.0	400
K ₂ HPO ₄	174.2	4.0	23
<u>Minor elements</u>			
FeCl ₃ ·6H ₂ O	270.3	0.54	2
H ₃ BO ₃	61.8	0.62	10
MnCl ₂ ·4H ₂ O	197.9	1.40	7
ZnCl ₂	136.3	0.10	0.7
Na ₂ EDTA	-	3.00	-
EDDHA (hydrogen ferric ethylene- diamine di-o-hydroxyphenyl- acetate)	-	1.17	-

Table 4. Continued

Compound	Molecular weight	Micrograms per liter	Nanomoles per liter
<u>Trace elements</u>			
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	237.9	5.0	21.0
$\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$	170.5	0.034	0.2
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	242.0	2.0	8.3
NH_4VO_3	117.0	2.3	19.7
$\text{K}[\text{Cr}(\text{SO}_4)_2] \cdot 12\text{H}_2\text{O}$	499.4	9.6	19.2
$\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$	262.9	4.5	17.1
$\text{Na}_2\text{WO}_4 \cdot 2\text{H}_2\text{O}$	329.9	1.8	5.5
$\text{Ti}_2(\text{C}_2\text{O}_4)_3 \cdot 10\text{H}_2\text{O}$	540.0	5.6	10.4
$\text{Al}_2(\text{SO}_4)_3$	342.2	3.2	9.4
As_2O_3	197.8	0.7	3.5
CdCl_2	183.3	0.8	4.4
SrSO_4	183.7	1.0	5.5
HgCl_2	271.5	0.7	2.6
PbCl_2	278.1	0.7	2.5
LiCl	42.4	3.1	73.1
Rb_2SO_4	267.0	0.8	3.0
NaBr	102.9	0.6	5.8
KI	166.0	0.7	4.2
NaF	42.0	1.1	21.2
Na_2SeO_4	188.9	1.2	6.4
$\text{Be}(\text{NO}_3)_2 \cdot 3\text{H}_2\text{O}$	187.1	10.4	55.6

Table 5. Saturation values for dissolved oxygen in Cache Valley, Utah: elevation 4700 feet, mean barometric pressure 0.85 atmosphere

Temperature (°C)	Saturation (mg/l)	Temperature (°C)	Saturation (mg/l)
0.0	12.03	14.0	8.48
0.5	11.87	14.5	8.39
1.0	11.70	15.0	8.30
1.5	11.55	15.5	8.21
2.0	11.39	16.0	8.13
2.5	11.24	16.5	8.04
3.0	11.09	17.0	7.96
3.5	10.94	17.5	7.89
4.0	10.80	18.0	7.80
4.5	10.66	18.5	7.74
5.0	10.51	19.0	7.66
5.5	10.39	19.5	7.59
6.0	10.25	20.0	7.51
6.5	10.12	20.5	7.45
7.0	10.00	21.0	7.38
7.5	9.87	21.5	7.32
8.0	9.75	22.0	7.25
8.5	9.63	22.5	7.19
9.0	9.51	23.0	7.12
9.5	9.40	23.5	7.07
10.0	9.28	24.0	7.01
10.5	9.18	24.5	6.95
11.0	9.07	25.0	6.89
11.5	8.97	25.5	6.84
12.0	8.87	26.0	6.79
12.5	8.76	26.5	6.73
13.0	8.67	27.0	6.68
13.5	8.58	27.5	6.64

VITA

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